sex cells of *rustica* var. *pumila* and *Bigelovii* var. *Wallacei* were grown last summer. There was no evidence of variation in the latter and no appreciable reduction in fertility. In the case of *rustica* there were both dwarfs and plants of normal size, but little decrease in fertility in the former. The treatment was apparently gametically or zygotically lethal for *suaveolens* and almost so in the case of *nudicaulis*. The former produced non-viable seeds only, and from seed of the latter species only two plants grew to maturity. One of them was normal in appearance and in flower, while the other was decidedly dwarf and set no seed until late in the season and then only in cleistogamous flowers. It would seem probable that progenies from both variant and "normal" plants of the two X-rayings of *tabacum* will exhibit recessive modifications, as may progenies from the other species which received similar treatment. It is proposed as soon as possible to study the effects of variations in X-ray dosage on the nature of the heritable variations which we now know can be obtained, and possibly to determine comparative effects of X-ray treatment of seeds of species in which variation has already been produced by treatment of sex cells. A complete account of the cytology and genetics of the X-ray variants will appear in the *Botanical Gazette*.

* A tall, large-leaved, red-flowered variety of the species of commerce, grown in the pure line for many years and extensively studied here in inter- and intraspecific hybrids (cf. *Univ. Calif. Publ. Botany*, vols. 5 and 11, and elsewhere).

GENETIC EFFECTS OF X-RAYS IN MAIZE

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Crossing-Over.—The frequency of crossing-over in the *C-Sh-Wx* region in maize, though varying rather widely in different plants, is remarkably constant within the individual. Repeated pollinations from the same heterozygous plant throughout the pollen-shedding period show no significant variation in cross-over percentage, indicating (for the male germ cells) that considerable differences in age and wide fluctuation in temperature and other weather conditions have no appreciable effect on crossing-over in this region.[2] The discovery by Mavor[3] that cross-over frequency in Drosophila is affected by X-rays even in the sex chromosome (in which Plough[4] and Bridges[5] had found crossing-over unaffected by age and temperature differences) suggested that cross-over variations in the *C-Sh-Wx* region might be induced by X-ray treatment. Later experiments by Muller[6] indicated that certain regions in Drosophila are relatively immune to the effects of X-rays on crossing-over.

In 1925, 40 plants of three families heterozygous for *C-Sh-Wx* were treated by X-raying the young tassels at the time of the maturation divisions. The stage in the development of the germ cells was determined by cytological examination of several representative plants in each family, and the treatments were applied at about the middle of the maturation period. A study of the course of maturation in several untreated plants, made by daily examination of anthers in aceto-carmine smears, had shown that the reduction divisions in the main tassel occur mostly during a period of five or six days, and that they follow a regular course through the inflorescence, so that the stage of development may be fairly accurately estimated by a single examination at any time during the period. In order to increase the proportion of germ cells exposed to treatment at any susceptible stage, three of the four series of treatments were applied intermittently, in three exposures at 48-hour intervals.

The treatments were applied in the open field, by means of a General Electric Portable X-ray Outfit, with a 10 milliampere self-rectifyng Coolidge tube. This machine is designed to operate at 88,000 volts (peak) at a line voltage of 110–112. The voltage in our experiments was somewhat lower, since the line voltage approximated 104.[7] All treatments were given with a current of $2^3/_4$ m. a. passing through the tube, and at a target distance of 20 cm., measured to the center of the irradiated tassel. The treatments were applied to four series of 10 plants each, as follows:

SERIES	TREATMENT
I	1 exposure, 5 minutes
II	3 exposures, 2 minutes each, at 48-hour intervals
III	3 exposures, 5 minutes each, at 48-hour intervals
IV	3 exposures, 8 minutes each, at 48-hour intervals

Microscopic examination of daily pollen specimens showed that these treatments were severe enough to kill an appreciable proportion of the pollen. The pollen of untreated tassels usually contained at least 95% of good, well-filled grains, while that of the treated tassels, particularly those given the heavier treatments, contained a considerable proportion of empty or defective grains, reaching more than 50% in extreme instances. In many cases the injury was apparent during only a part of the pollen-shedding period, good pollen being produced both before and after. In the tassels receiving the two heavier treatments the number of anthers shedding pollen also was materially reduced. The untreated tassels of treated plants regularly produced good pollen.

The cross-over percentages in male gametes in treated and untreated tassels are shown in table 1. These data show some small but apparently significant differences in cross-over frequency. Treatment II shows a significantly lower percentage of *Sh-Wx* cross-over than the check in family

5009 and a significantly higher percentage in family 5017, and treatment I shows a significantly higher percentage in family 5001.

It may be shown, however, that these differences are not ascribable to the X-ray treatment. Since different plants within a family vary rather widely in crossing-over, considerable differences between small groups of treated and untreated plants may result from the accidental choice for treatment of plants normally high or low in crossing-over. In other words, there is a sampling error due to variation between plants, which is not measured in the probable errors determined from totalled progenies. The error in sampling plants may be avoided, however, by determining the effect of treatment only in comparisons of treated and untreated tassels

TABLE 1

EFFECT OF X-RAY TREATMENTS ON CROSSING-OVER

	CROSS-OVER PERCENTAGE			
	TREATMENT		UNTREATED	
TREATMENT	C-SH	SH-WX	C-SH	SH-WX
I (Family 5001)	4.33±0.24	23.84±0.27	4.69±0.24	21.80±0.47
II	2.87±0.24	14.81±0.48	3.92±0.20	17.15±0.38
III	3.70±0.26	17.49±0.50		
IV (Family 5009)	2.12±0.30	16.05±0.58		
II	4.25±0.23	24.27±0.49	4.00±0.13	20.95±0.28
III	3.95±0.26	21.57±0.54		
IV (Family 5017)	3.81±0.26	22.59±0.54		

of the same plants, for the earlier work has shown that different tassels of the same plant do not differ appreciably in cross-over frequency.

In 16 of the treated plants pollinations were made from both treated and untreated tassels. The difference in cross-over percentage was less than 3 times its probable error in all but 2 of the 16 cases for the *Sh-Wx* region, and in all but one case for the *C-Sh* region. In most of these cases daily pollinations were made in a search for effects of treatment which might last for only a part of the shedding period, but the results showed no grouping of high or low variations on successive days. These trials, therefore, show no appreciable effect of X-rays on cross-over frequency.

However, since the three "significant" differences just mentioned all occurred among the plants given the heaviest treatment, and since these three plants were slightly more advanced at the time of treatment than most of the other treated plants, a trial with heavier dosage and one more fully covering the maturation period seemed desirable. Accordingly, a second trial was made, in which the X-ray treatments were applied to the tiller tassels (which pass through the maturation stage much more quickly)

in daily applications beginning before maturation and continuing until after its completion. Four plants were given six exposures of five minutes each, four plants eight exposures of five minutes each, and two plants eight exposures of ten minutes each. Voltage, milliamperage and target distance were the same as before. In order to secure data for another chromosome region in the same trial, stocks were prepared heterozygous for *Sh-Wx* and also for *Y-Bh*, the only other endosperm linkage now available in maize. Unfortunately, it was found that in the stocks used the aleurone blotch *Bh* could not be distinguished accurately enough on starchy grains to permit critical determinations. The data from this trial, therefore, refer only to the *Sh-Wx* region.

The heavy treatments used in this trial greatly reduced the yield and viability of pollen in all cases. Only one treated tassel shed pollen more than two days. The amount shed was in most cases too small to permit the pollination of more than one or two ears per day. Microscopic examination of the pollen shed showed a very high proportion of defective grains, often higher than 50% and in some cases higher than 90%. The tassels given eight exposures of ten minutes each shed no pollen.

In seven of the ten plants treated, it was possible to determine cross-over frequency in both treated and untreated tassels. In each of these cases the difference in cross-over percentage was less than three times its probable error. The results give no indication of any effect of the X-ray treatment used on crossing-over in this region, or of any differential elimination of cross-over gametes by lethal doses of X-rays.

Chromosome Deficiency.—When plants recessive for an endosperm character are pollinated by the corresponding dominant, the heterozygous seeds produced include occasional mosaic individuals, in which a portion of the endosperm shows the recessive character. Emerson[8] has shown by genetic evidence that this is due, at least in large part, to aberrant chromosome behavior, since unlinked genes are lost independently while linked genes are almost always lost together. Because of the relative rarity of the phenomenon it has not been studied cytologically, and the nature of the process causing the deficiency is unknown. Non-disjunction and chromosome elimination fit the observed facts equally well. It is possible that gene mutation accounts for the small proportion of cases in which linked genes appear to be lost independently, but there are several possible explanations not yet excluded which may account for these cases without gene mutation.

To determine the effect of X-rays on this phenomenon the ears in which the frequency of mosaic endosperms was to be determined were X-rayed at or shortly before the time of fertilization, which, according to various observers, normally occurs about 24–28 hours after pollination. Voltage, milliamperage and target distance were the same as in the experiments on

TABLE 2

EFFECT OF X-RAY TREATMENTS ON THE FREQUENCY OF OCCURRENCE OF MOSAIC ENDOSPERMS

TRIAL	TREATMENT: NUMBER OF EXPOSURES	DURATION OF EXPOSURES MIN.	TOTAL TIME OF EXPOSURES MIN.	INTERVAL BETWEEN EXPOSURES HRS.	PERIOD FROM POLLINATION TO EXPOSURES HRS.		FREQUENCY OF MOSAICS* FOR CHARACTERS OF CHROMOSOME — I (*C Wx, I*)	II (*R*)	III (*Su*)	V (*Y*)	VIII (*Pr*)	TOTAL FREQUENCY	TOTAL PERCENTAGE
I	1	5	5	–	24	Treated	1/135	0/122	2/417	1/245	0/58	4/977	0.41
						Untreated	0/273		0/294	0/185		0/752	0
	2	5	10	2	24–26	Treated		7/153	5/165	1/12	0/71	13/401	3.24
						Untreated	2/451	1/85	1/499	0/451	0/43	4/1529	0.26
II	3	10	30	3	21–27	Treated		19/314	16/447		0/132	35/893	3.92
						Untreated		2/599	1/675		1/336	4/1610	0.25
III	4	5	20	2	22–28	Treated	6/90	38/414	47/831		4/143	95/1478	6.43
						Untreated		3/507	0/703		0/126	3/1336	0.22
IV	5	5	25	2	20–28	Treated		18/262	8/297		8/297	34/856	3.97
						Untreated		0/385	1/395		0/174	1/954	0.10
Total			Frequency			Treated	7/225	82/1265	78/2157	2/257	12/701	181/4605	
						Untreated	2/724	6/1576	3/2566	0/636	1/679	12/6181	
			Percentage			Treated	3.11	6.48	3.62	0.78	1.71	3.93	
						Untreated	0.28	0.38	0.12	0	0.15	0.19	

* Including only mosaics in which the recessive character covered at least $^1/_8$ of the surface. The frequency is stated for each chromosome as total number of mosaics observed/total number of seeds in which a mosaic for this chromosome could have been detected.

crossing-over. In most cases intermittent exposures were given. Details of treatment are shown in table 2, together with the summarized results.

The four trials are not directly comparable, since they were made on different days and since several families were used in order to secure data for different chromosomes. Within each trial the treated and untreated groups were pollinated at the same time and from the same sample of pollen, and in most though not in all cases they include only ears of the same families. In determining the results mosaic endosperms showing the recessive characters on approximately one-half, one-quarter and one-eighth of the surface were recorded separately, since these presumably represent losses occurring at or before the first, second and third divisions, respectively. These have been summarized in preparing the table. Smaller spots were not included, since they cannot always be identified with certainty.

The results show a pronounced increase in the percentage of mosaic endosperms following X-ray treatment. It is possible that this effect may be due in part to an increase in the frequency of gene mutations as well as that of chromosome aberrations. The immense effect of X-rays on gene mutation in Drosophila, as recently reported by Muller,[9] suggests that this may be a factor. But unless the normal frequency of mutation during endosperm development is very much higher than that of the same genes previous to maturation,[10] even such extreme increases by X-ray treatment as those reported by Muller would not produce enough gene mutations to affect materially the percentage of mosaic endosperms. Unfortunately, direct evidence of the chromosomal nature of mosaics for chromosomes II, III and VIII, whether in treated or untreated material, cannot be obtained until linkages of endosperm genes in these chromosomes are found. Such evidence is obtainable for chromosomes I and V, but in these trials only a few mosaics for these chromosomes were included. The majority of these involved linked characters, and in all cases the linked genes were lost together, indicating that the mosaics in these cases at least were chromosomal.

The percentage of mosaics in the X-rayed series as a whole is more than 20 times as great as that in the untreated series. The frequency of mosaics for the five linkage groups varies somewhat, but since the data were obtained in part from different families, the significance of the differences is doubtful. With so high a frequency of aberrant divisions cases involving more than one chromosome would be expected, resulting in the simultaneous loss of unlinked genes. One such case was found in which the unlinked genes *I* and *Su* apparently were lost in the first division.

The frequency of the larger mosaics was increased by treatment more extremely than that of the smaller, indicating a rather rapid decrease in the effect of radiation. The frequency of "first division mosaics" was

2.56% in X-rayed seeds and 0.08% in untreated seeds. If these average frequencies, determined from five paternal chromosomes (but chiefly from chromosomes II and III), may be taken to represent roughly the frequency of loss of the 30 chromosomes present in the triploid endosperm nucleus, an average rate of about 75 aberrancies per 100 mitoses is indicated for the first division in endosperm development in the irradiated seeds.

[1] Formerly NATIONAL RESEARCH FELLOW. Part of the work reported was done under this fellowship at the Bussey Institution, Harvard University, 1925–26.

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GENERAL THEORY OF POLYGENIC OR NON-MONOGENIC FUNCTIONS. THE DERIVATIVE CONGRUENCE OF CIRCLES

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This paper is devoted mainly to the geometric questions connected with the differentiation of a general complex function $w = \varphi(x, y) + i\psi(x, y)$ with respect to the independent variable $z = x + iy$. We assume merely that the components φ and ψ are continuous and have continuous partial derivatives in the region considered, but do not assume the Cauchy-Riemann equations.

The limit of the increment ratio $\Delta w/\Delta z$, in general, then depends not only on the point $x + iy$ but also on the direction θ (or the slope m) along which the neighboring point approaches the given point. Thus the derivative dw/dz has, in general, infinitely many complex values at a given point. We call such a function *polygenic*. Only when the Cauchy-Riemann equations are fulfilled will the derivative have a unique value at the point; the function is then called *monogenic*. Thus the derivative of a polygenic function is a function of z and m while the derivative of a monogenic function is a function of z alone.